

Product datasheet

Human Methotrexate ELISA Kit ab213976

1 Image

Overview

Product name Human Methotrexate ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
1	20	265.2ng/ml		9.8%
2	20	4.5ng/ml		20.5%

Inter-assay

Sample	n	Mean	SD	CV%
1	10	298.9ng/ml		17.1%
2	10	5.2ng/ml		22.4%

Sample type Urine, Serum, Plasma

Assay type Indirect

Sensitivity 0.087 ng/ml

Range 0.13 ng/ml - 1000 ng/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Urine	122.67	100% - 161%
Serum	82	71% - 86%
Plasma	91.67	77% - 107%

Assay time 1h 30m

Assay duration Multiple steps standard assay

Species reactivity**Reacts with:** Mouse, Rat, Human**Product overview**

The Human Methotrexate ELISA Kit (ab213976) is a complete kit for the quantitative determination of methotrexate in serum, plasma and urine samples. Please read the complete kit insert before performing this assay. The methotrexate ELISA uses a methotrexate polyclonal antibody to bind methotrexate in the sample or standard competitively to that pre-bound to the wells as a bovine serum albumin (BSA) conjugate. Anti-methotrexate antibody bound to methotrexate in the sample or standard are washed away while those captured by the immobilized methotrexate are detected with a secondary antibody horseradish peroxidase (HRP) conjugate. The assay is developed with tetramethylbenzidine (TMB) substrate and the resulting absorbance is measured with a microplate reader at 450nm. The intensity of the yellow color is inversely proportional to the concentration of methotrexate.

Notes

Methotrexate is a drug used in the treatment of cancer and autoimmune disease. It is designed as an anti-folate to inhibit the metabolism of folic acid. Two distinct mechanisms of action have been described for methotrexate. In cancer treatments, methotrexate competitively inhibits the dihydrofolate reductase (DHFR) by blocking folate binding. DHFR converts dihydrofolate to active tetrahydrofolate. Inhibition of DHFR results in inhibition of the synthesis of purine and pyrimidine bases effectively limiting DNA and RNA synthesis and cancer cell growth. In autoimmune disease and specifically in the treatment of rheumatoid arthritis, methotrexate appears to impact several pathways resulting in inhibition of T cell activation. The effects include suppression of T cell expression of intercellular adhesion molecules, inhibition of methyl transferase activity and increased CD95 sensitivity leading to apoptosis in active T cells.

Monitoring methotrexate levels is important to assure appropriate levels are maintained during therapy or treatment. High levels of methotrexate can lead to toxicity and potential renal failure as well as immunosuppression. Additionally, methotrexate is known to interact with a wide variety of drugs leading to additional complications. Determining the presence of methotrexate in samples from subjects in blinded research studies can assist in the interpretation of study results.

Methotrexate is established as one of the most effective and safe therapeutics for rheumatoid arthritis. The safety profile assures that methotrexate will continue to be used in new studies in combination with other new or established drugs. The same is true in its use as a cancer therapeutic. The Methotrexate ELISA enables monitoring levels of methotrexate in both preclinical and clinical research. The methotrexate assay is also appropriate for the detection of methotrexate contamination after its use as a selective agent for recombinant protein production in mammalian cell lines.

Platform

Pre-coated microplate (12 x 8 well strips)

Properties**Storage instructions**

Store at +4°C. Please refer to protocols.

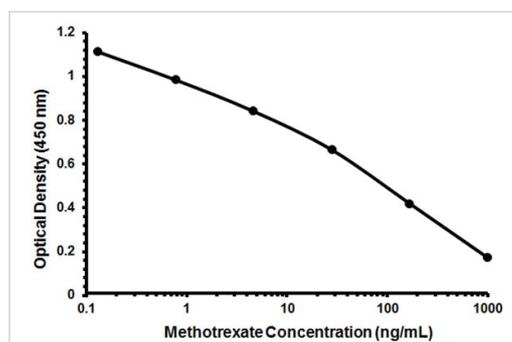
Components	1 x 96 tests
Antibody Diluent	1 x 6ml
Assay Buffer	1 x 50ml
Methotrexate Antibody (Lyophilized)	1 vial
Methotrexate coated microplate (12x 8 well strips)	1 unit

Components	1 x 96 tests
Methotrexate Conjugate	1 x 10ml
Methotrexate Standard (1,000ng)	2 vials
Plate Sealer	3 units
Stop Solution	1 x 10ml
TMB Substrate	1 x 10ml
Wash Buffer Concentrate	1 x 100ml

Relevance

Methotrexate (rINN) abbreviated MTX and formerly known as amethopterin, is an antimetabolite and antifolate drug used in treatment of cancer, autoimmune diseases and as an abortifacient in the induction of medical abortions. It acts by inhibiting the metabolism of folic acid. Methotrexate competitively inhibits dihydrofolate reductase (DHFR), an enzyme that participates in the tetrahydrofolate synthesis. The affinity of methotrexate for DHFR is about one thousand-fold that of folate for DHFR. Methotrexate acts specifically during DNA and RNA synthesis, and thus it is cytotoxic during the S-phase of the cell cycle. Logically, it therefore has a greater toxic effect on rapidly dividing cells which replicate their DNA more frequently, and thus inhibits the growth and proliferation of these non-cancerous cells. Lower doses of methotrexate have been shown to be very effective for the management of rheumatoid arthritis, Crohn's disease, and psoriasis.

Images



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Standard Curve.

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